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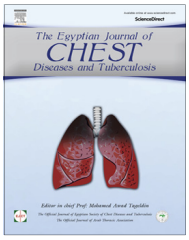

ORIGINAL ARTICLE

Significance of *Moraxella catarrhalis* as a causative organism of lower respiratory tract infections

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KEYWORDS

Respiratory infection;
M. catarrhalis;
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Abstract *Background:* *Moraxella catarrhalis* is an exclusively human pathogen that may be overlooked in culture because of its phenotypic similarity to commensal Neisseria. Indeed, beta lactamase producing isolates of *M. catarrhalis* appear to be wide spread, and this may play an important role in the therapy of infections, particularly in the treatment of mixed infections

Objective: The purpose of this study was to evaluate the significance of *M. catarrhalis* as a pathogen in causing lower respiratory tract infection.

Methods: This study was carried out on 200 patients who were diagnosed as having lower respiratory tract infection and admitted during the period of the research to chest unit of Tanta University hospitals, another 50 adult volunteer were considered as control group during the period from January 2014 to August 2014. All patients were subjected to the following assessment, full clinical history; their records were reviewed for name, age, sex, and special habits. Patients suspected to be suffering from lower respiratory tract infection were considered. Only sputum samples of high bacteriological quality were analyzed. All specimens were cultured.

Result: *M. catarrhalis* is responsible for 11.5% of all cases of lower respiratory tract infection included in this study. Infection occurs more common in patients having underlying lung disease especially chronic pulmonary diseases.

Conclusion: This study shows that when microbiological and clinical criteria are met, *M. catarrhalis* when isolated should be considered as a pathogen causing lower respiratory tract infections. *M. catarrhalis*, lower respiratory tract infections.

Introduction

Moraxella catarrhalis is a Gram negative, aerobic, diplococcus frequently found as a commensal of the upper respiratory tract. Colonies on blood agar are non-hemolytic, round, opaque, convex, and grayish white. The gram negative diplococcus

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is difficult to distinguish from Neisseria spp. in a typical Gram staining. Various biochemical test methods exist to distinguish the species. *M. catarrhalis* is DNAse, catalase and oxidase positive, furthermore the pathogen hydrolyzes tributyrin, reduces nitrate and is unable to produce acid from glucose, lactose, maltose fructose and sucrose. None of these tests are 100% sensitive or specific. The informative value of more sensitive DNA methods such as polymerase chain reaction (PCR) has been demonstrated [1].

Two major phylogenetic subpopulations (type 1 and type 2 strains) of the species have been identified. [2]. The virulence factors for *M. catarrhalis* include; Lipo-oligosaccharide (LOS) of the cell wall, complement resistance and the polysaccharide capsule [3].

The genus Moraxella

The genus Moraxella belongs to the family Moraxellaceae, which includes the closely related genera Acinetobacter and Psychrobacter. The Moraxella genus itself currently contains 15 different species. Although all these 16 species have been classified within the genus Moraxella, the classification of the Moraxellaceae family is still not definitive but continuously evolving. In adults the pharyngeal carriage rate is noticeably lower and varies between 1% and 5%. It increases again in adults older than 60 years of age [4].

Two distinct genetic lineages related to 3 different 16S rRNA types have been identified for *M. catarrhalis*, which differ phenotypically in their ability to resist the killing effect of human serum (sero-resistant versus sero-sensitive), and in their ability to adhere to human epithelial cells [5]. It is known that *M. catarrhalis* exists in biofilms and that it is able to prolong its survival through biofilm formation on mucosal surfaces in the nasopharynx [6]. Biofilm formation has been demonstrated for numerous pathogens and is clearly an important microbial survival strategy [7]. Furthermore, several nosocomial outbreaks of respiratory disease due to *M. catarrhalis* infections in adults and in children have been reported [8]. In children, *M. catarrhalis* causes mainly upper respiratory tract infections (otitis media), whereas in adults the pathogen causes lower respiratory tract infections in previously compromised airways (acute exacerbation of Chronic Obstructive Pulmonary Disease [COPD]). Invasive infections such as bacteraemia, meningitis, septic arthritis, ventriculitis and endocarditis, are very rare [9].

Of added importance, and perhaps contributing to the apparent increase in *M. catarrhalis* as a respiratory tract pathogen, has been the production of beta-lactamase enzymes. There has been rapid acquisition and spread of beta lactam antibiotic resistance of *M. catarrhalis* in the last 20–30 years to the extent that approximately 95–99% of clinical isolates now appear to resist one or more beta lactam antibiotics [10].

Aim of the work

The aim of this study was to evaluate the significance of *M. catarrhalis* as a pathogen in causing lower respiratory tract infection, characterize the cell surface hydrophobicity and beta-lactamase production of *M. catarrhalis* as important virulent factors (see Figs. 1–4).

Materials and methods

The present study was carried out in Microbiology and Immunology Department, Faculty of Medicine, Tanta University, on 200 patients who were clinically diagnosed as having lower respiratory tract infection and admitted in the period from January 2014 to August 2014 to chest unit of Tanta University hospitals. Full clinical history was taken from the selected patients and their records were reviewed for name, age, sex, special habits, clinical diagnosis on admission, clinical signs of infection and immune suppression status e.g. steroid therapy, malignancy or diabetes mellitus. Patients with immune suppression status were excluded. The most common primary underlying disease was Chronic Obstructive Pulmonary Disease (COPD), in addition to bronchiectasis and pulmonary fibrosis. Another 50 (29 male and 21 female) apparently healthy volunteers with no symptoms and signs suggesting respiratory tract infection were included in this study as a control group. The age was ranging from 20 to 73 years (mean age 57). History of smoking was present in 17 individual.

A. Sample collection and transport: Sputum samples: Early morning sputum samples were obtained from 200 patients. Every patient was instructed to brush his or her teeth and to gargle with water immediately before obtaining the sputum sample to reduce the number of contaminations with oropharyngeal bacteria [12].

Throat swabs

Throat swabs were obtained from healthy control subjects. A bright light from over the shoulder of the specimen collector was focused into the oral cavity so that the swab can be guided to the posterior pharynx [12].

Processing of specimens:

(1) *Microscopy:* smears were prepared from the most purulent part of the sputum samples and stained with Gram stain [13]. Only sputum samples of high bacteriological quality were analyzed (<10 epithelial cells, >25 leucocytes/low power field) [14].
(2) *Culture:* all specimens were inoculated on blood, chocolate, McConkey, and *Sabouraud* agar. The inoculated blood and chocolate plates were incubated at 37 °C for 24 h in candle jar containing 3–5% Co_2.

The following criteria were considered for determining pathogenic significance of an isolate [14]:

(1) Clinical evidence of infection consistent with the disease spectrum associated with *M. catarrhalis* (cough with sputum production).
(2) *M. catarrhalis* as a predominant potential pathogen isolated from an appropriate and adequate specimen.
(3) Clinical response on treatment with antibiotic to which the isolate was susceptible.

Taking the above mentioned criteria into consideration, as follows:-

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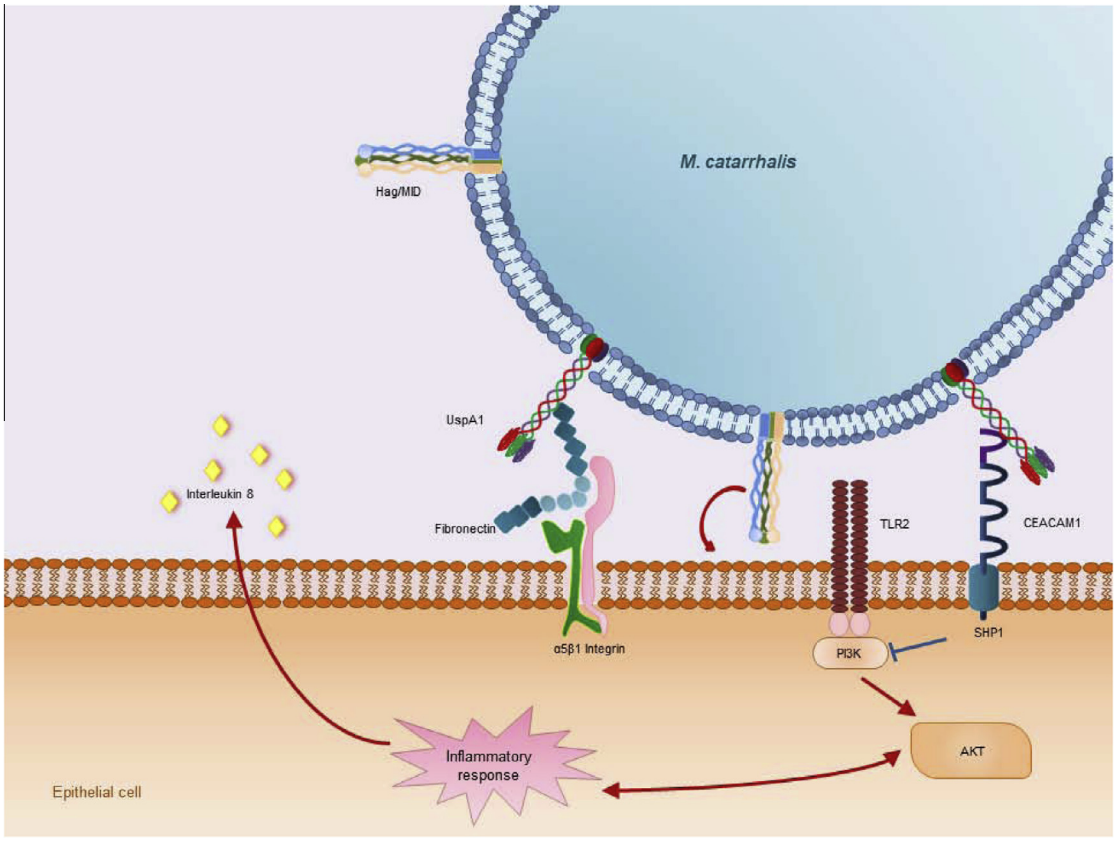

Figure 1 Adherence to host epithelial cells [11].

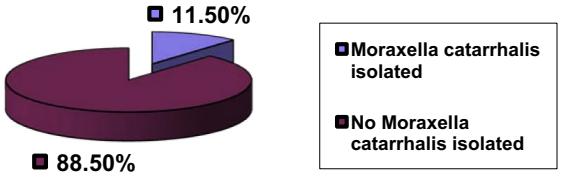

Figure 2 Prevalance of *Moraxella catarrhalis* isolation in patient group.

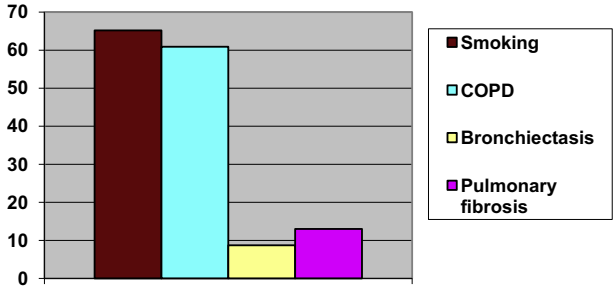

Figure 3 Primary underlying diseases.

- *Significant isolate:* if criteria 1, 2 and 3 were present.
- *Probably significant isolate:* if criteria 1 and 2 were present and 3 could not be assessed.
- *Indeterminate isolate:* if only criterion 1 was present.

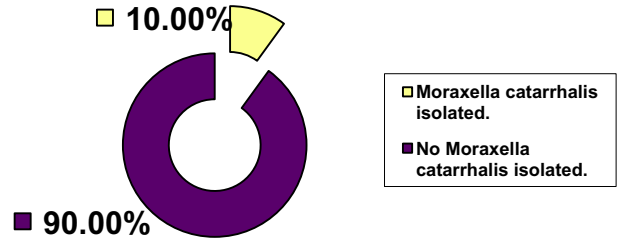

Figure 4 Prevalence of *Moraxella catarrhalis* isolation in the control group.

Results

This work was carried out in the microbiology and immunology department, faculty of medicine, Tanta University in the period from January 2014 to August 2014. This study comprised 200 patients who were clinically diagnosed as having lower respiratory tract infection and admitted during the period of the research to the chest unit of Tanta University hospitals. Another 50 apparently healthy volunteers with no symptoms and signs suggesting respiratory tract infection were also included in this study as a control group. The results of this study were collected analyzed and tabulated as follows (see Tables 1–8).

Discussion

M. catarrhalis is an exclusively human commensal and mucosal pathogen. Its role as a disease-causing organism has long been questioned [15]. In general, the organism is mostly associated with upper respiratory tract infections especially in children and lower respiratory tract infections in adults especially exacerbations of Chronic Obstructive Pulmonary Disease (COPD). [11]. This study was performed on the sputa of 200 patients who were clinically diagnosed as having lower respiratory tract infection, in addition to another 50 throat swabs from 50 adult healthy volunteers as a control group. The prevalence of *M. catarrhalis* isolation in patient group was 11.5%. This result is not far from that of Mackenzie et al. [16]who reported that 17.1% of adult patients with respiratory tract infections were due to *M. catarrhalis*. In early reports the recognition of *M. catarrhalis* as a human lower respiratory tract pathogen has been delayed for several reasons. One of these reasons is that the organism was considered as an upper respiratory tract commensal [17].

In this study 69.6% of the patient isolates were from patients aged over 50 years. Anita et al. [18] reported a similar percentage which was 68% from the same age group.

In this study they only focused on pure cultures which explain the lower percentage of *M. catarrhalis* isolation rate in this age group. The Prevalence of *M. catarrhalis* isolation in our control group was 10%. Of that 2% was isolated from control subjects aged from 20 to 50 years and 8% was isolated from control subjects aged over 50 years. A percentage of 11.7% for control subjects aged over 50 years was reported by Sehgal and Al Shaimy [19], which is close to our result.

The most common isolated organisms other than *M. catarrhalis* in our patient group were *Streptococcus pneumoniae*, *Klebsiella pneumoniae* and *Staphylococcus aureus*.

In the patient group 60.9% of our isolate were from males and 39.1% were from females. Tamang et al. [13] reported similar male to female ratio which was 64% from male and 36% from female patient. A higher male ratio is probably due to the habit of smoking, which is more common among males.

65.2% of *M. catarrhalis* isolates in the patient group were in winter months. Similar data were reported by George et al. [20] who found that infection with *M. catarrhalis* was more common in winter months. The organism was isolated from control subjects in winter months in a significant higher rate than spring and summer months (P-value < 0.001). A rate which agrees with the result was reported by Hendley et al. [21].

History of smoking was present in 65.2% of the patient group. Similar finding was reported by Nicolas et al. [22] and 1991. Kurtti et al. [23]described that the antibody titers against *M. catarrhalis* were significantly higher in smokers than non smokers.

Table 1 Age distribution of the patient group.

Age group	No. of patients (n = 200)	%
20–50 year	116	58
> 50 year	84	42

Table 2 Sex distribution of the patient group.

Sex	No. of patients (n = 200)	%
Male	123	61.5
Female	77	38.5

Table 3 Percentage of different clinical findings in the patient group.

Clinical findings	No. of patients (n = 200)	%
Purulent sputum	138	69
Fever	113	56.5
Wheezes	87	43.5
Haemoptysis	17	8.5
Positive findings in chest X-ray	144	72

Table 4 Prevalence of different organisms' isolation in the patient group.

Organism	No. of isolates (n = 200)	%
Streptococcus pneumoniae	34	17
Candia species	17	8.5
Acinetobacter species	17	8.5
Kingella kingae	2	1
Moraxella catarrhalis	23	11.5
Klebsiella spp.	22	11
Staphylococcus aureus	22	11
Pseudomonas aeruginosa	21	10.5
Haemophilus influenza	1	0.5
Streptococcus pyogenes	10	5
Neisseria meningitidis	1	0.5
E. coli	4	2
Stenotrophomonas maltophilia	1	0.5
Commensals	20	10
No growth	5	2.5

Table 5 Percentage of age related *Moraxella catarrhalis* isolates to the total number of *Moraxella catarrhalis* isolated in the patient group.

Age group	No. of isolates (n = 23)	% to the total isolates	
20–50	7	30.4	Chi-Sq = 7.043
> 50	16	69.6	P-Value = 0.008

COPD was identified as a risk factor for development of *M. catarrhalis* lower respiratory tract infection. In our patient group COPD cases represent 60.9% of all cases of *M. catarrhalis* infection. A much higher incidence 76% was observed by Ahmed et al. [24]. A much lower incidence 37% was reported by Anita et al. [18].

Antibiotic susceptibility testing revealed that 91.3% of the strains isolated from patient group were resistant to Penicillin. Surveys of resistance in the UK in 1991 showed that 91% of *M. catarrhalis* strains, respectively, were resistant to Penicillin.

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Table 6 Distribution of *Moraxella catarrhalis* isolation in patient group over months of the year (2014).

Month	No. of isolates (n = 23)	%
January	8	34.8
February	4	17.4
March	3	13
April	4	17.4
May	3	13
June	1	4.3
July	0	0
August	0	0

Table 7 Antibiotic susceptibility of *Moraxella catarrhalis* isolated from the patient group.

Antibiotic	*Moraxella catarrhalis* (n = 23)			
	Sensitive		Resistant	
	No. of isolates	%	No. of isolates	%
Penicillin (10 μg)	2	8.7	21	91.3
Amoxicillin–clavulanic acid (20 μg)	23	100	0	0
Cefaclor (5 μg)	22	95.7	1	4.3
Cefuroxime (30 μg)	23	100	0	0
Ceftriaxone (30 μg)	23	100	0	0
Ciprofloxacin (5 μg)	23	100	0	0
Erythromycin (15 μg)	23	100	0	0
Tetracycline (30 μg)	22	95.7	1	4.3
Gentamycin (10 μg)	23	100	0	0
Co-trimoxazole (25 μg)	23	100	0	0

Table 8 Beta-lactamase production and hydrophobicity of *Moraxella catarrhalis* isolates in patient and control groups.

	Beta-lactamase production				Hydrophobicity			
	Positive		Negative		Hydrophobic		Hydrophillic	
	No.	%	No.	%	No.	%	No.	%
Patient group Isolates (n = 23)	22	95.7	1	4.3	18	78.3	5	21.7
Control group Isolates (n = 5)	4	80	1	20	1	20	4	80

An important factor in the pathogenesis of numerous diseases is hydrophobicity. It is related to the adhesive ability of the organism, adhesive ability increases as the cell surface hydrophobicity increases and decreases as the cell surface hydrophobicity decreases [25]. Janicka et al. [25]found that hydrophobic strains of *M. catarrhalis* were isolated more often from sputum rather than from nose and throat swabs, and that hydrophillic strains were found in statistically significant incidence in samples from upper (48.7%) than lower (10.5%) respiratory tract.

Conclusions

From the present study, it could be concluded that *M. catarrhalis* is responsible for 11.5% of all cases of lower respiratory tract infection in the study. Infection and colonization occur more commonly in elderly patients. Infection occurs more common in patients having underlying chronic pulmonary diseases and in smokers. Most of the pathogenic strains were beta-lactamase producers and hydrophobic and most of the pathogenic strains were susceptible to all tested antibiotics.

Conflict of interest

There is no conflict of interest.

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